

## The problems of living in the sea: the uptake of inorganic carbon and nutrients in *Posidonia oceanica* (L.) Delile.

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The genus *Posidonia* exhibits a peculiar geographical distribution. It is composed by nine species, eight of which are distributed along the Australian coasts and only one, *Posidonia oceanica* (L.) Delile, is a Mediterranean endemism. Like other angiosperms, *P. oceanica* has adapted secondarily to the marine environment, and has developed anew mechanisms to face a liquid and alkaline medium (pH 8.2) that contains a high salt concentration (0.5 M NaCl). The liquid environment limits the diffusive flow of CO<sub>2</sub> and nutrients and, furthermore, CO<sub>2</sub> dissolves in water and forms HCO<sub>3</sub><sup>-</sup>, the more abundant chemical species of inorganic carbon at pH 8.2.

Like other green plants *P. oceanica* uses CO<sub>2</sub> for photosynthesis. In addition, this species shows a transport system in the plasma membrane for the direct uptake of HCO<sub>3</sub><sup>-</sup>, that uses H<sup>+</sup> as the driving ion. The addition of HCO<sub>3</sub><sup>-</sup> provokes a transient hyperpolarization of the plasma membrane followed by a depolarization; at the same time, the cytosolic pH (pH<sub>c</sub>) becomes transiently acidic and next it gets alkaline, and remains alkaline throughout the HCO<sub>3</sub><sup>-</sup> pulse. The alkalization of the pH<sub>c</sub> is due to the cytosolic accumulation of HCO<sub>3</sub><sup>-</sup> and OH<sup>-</sup> and it is sensitive to the addition of ethoxymylamide, an inhibitor of the internal carbonic anhydrase. The increase of negative charges in the cytosol triggers the release of Cl<sup>-</sup> to recover the values of the resting membrane potential. The plasmalemma of *P. oceanica* exhibits a reduced Na<sup>+</sup> permeability and shows a H<sup>+</sup>/Na<sup>+</sup> antiporter activity that keeps low and relatively constant the cytosolic Na<sup>+</sup> concentration (17 mM Na<sup>+</sup>). The inside negative membrane potential (-178 mV) and the low [Na<sup>+</sup>]<sub>c</sub> generate a tremendous Na<sup>+</sup>-motive force that this plant uses for the high affinity transport of NO<sub>3</sub><sup>-</sup> (Km= 21 μM), and of the amino acids alanine (Km= 37 μM) and cysteine (Km= 10 μM). The uptake of these compounds shows a strict dependence on the presence of Na<sup>+</sup> in the medium. Moreover, the addition of micromolar concentrations of NO<sub>3</sub><sup>-</sup>, alanine or cysteine gives rise to millimolar increments of [Na<sup>+</sup>]<sub>c</sub>. Experiments with external LIX pH mini-electrodes show that the uptake of glucose is not Na<sup>+</sup> but H<sup>+</sup> dependent. Thus, the model for the ion transport energization in this species seems to be mixed, with a H<sup>+</sup>-ATPase as the primary pump and a series of carriers that use H<sup>+</sup> (HCO<sub>3</sub><sup>-</sup>, Na<sup>+</sup>, glucose) or Na<sup>+</sup> (NO<sub>3</sub><sup>-</sup>, amino acids) as the driving ion.

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